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Review Article

Physiological changes due to hepatotoxicity and the protective role of some medicinal plants

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ABSTRACT

The liver is the largest, important organ and the site for essential biochemical reactions in the human body. It has the function to detoxify toxic substances and synthesize useful biomolecules. Therefore, damage to the liver leads to grave consequences. This damage resulted from chronic alcoholic abuse, viral hepatitis or inherited metabolic disease. Liver damage is associated with cellular necrosis, fibrosis, and increase in tissue lipid peroxidation and depletion in tissue glutathione level. Most of the hepatotoxic chemicals damage liver cells mainly by inducing lipid peroxidation and other oxidative damages in the liver. Natural antioxidants are found in many compounds classified as secondary plant metabolites, e.g. polyphenols (phenolic acids and flavonoids) and terpenoids (carotenoids), and the consumption of foods that contain these compounds in large quantities seems to play an important role in prophylaxis against many diseases. Herbal medicines derived from plant extracts are being increasingly utilized to treat a wide variety of clinical disease. More attention has been paid to the protective effects of natural antioxidants against drug induced toxicities especially whenever free radical generation is involved. Popularity of herbal remedies is increasing and at least one quarter of patients with liver disease use botanicals. The World Health Organization (WHO) estimates that 80 percent of the population of some Asian and African countries presently use herbal medicine for some aspect of primary health care. Some medicinal herbs have proven hepatoprotective potential. *Silybum marianum* (milk thistle) has been used to treat liver diseases since the 16th century. Its major constituents are the flavonoids silibinin, silydianin, silychristin, and isosilibinin, of which silibinin is the biologically most active compound and used for standardization of pharmaceutical products.

1. Introduction

The liver is one of the largest organs in the human body and the chief site for intense metabolism and excretion (Ahsan et al., 2009). It plays a major role in detoxification and excretion of many endogenous and exogenous compounds; any injury to it or impairment of its functions may lead to many implications on one's health (Subramaniam et al., 2015). Hepatic damage is associated with distortion of these metabolic

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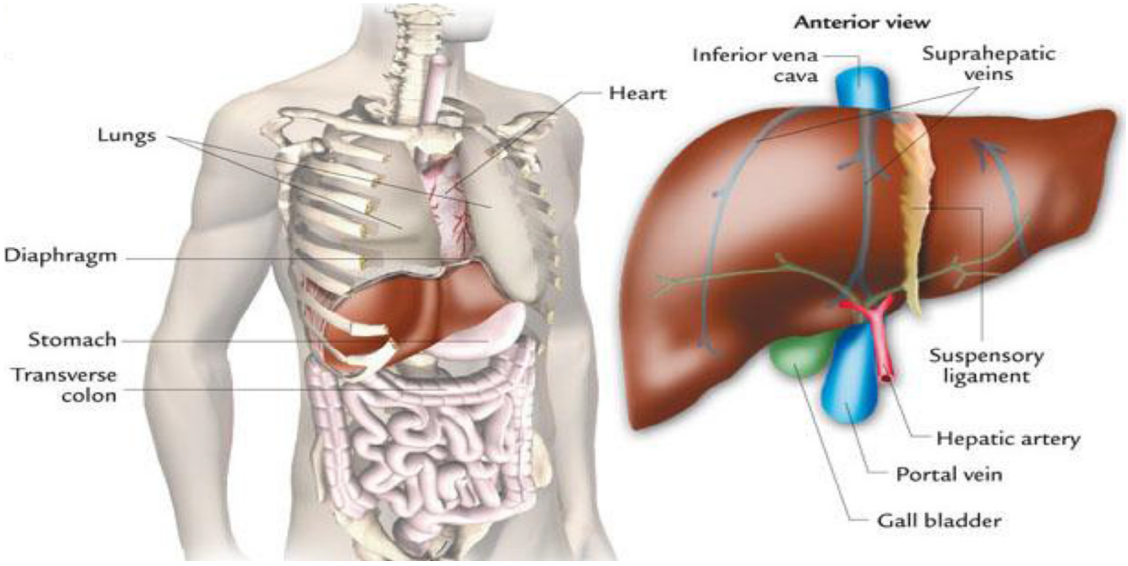


Fig. 1 – The liver (Polaxa, 2015).

functions. Liver damage is associated with cellular necrosis, increase in tissue lipid peroxidation and depletion of reduced glutathione levels. In addition, serum levels of many biochemical markers like transaminases, alkaline phosphatase, bilirubin, triglycerides and cholesterol are elevated in liver disease (Subramaniam et al., 2015). Liver diseases pose a serious challenge to international public health (Ahsan et al., 2009).

The liver plays a central role in transforming and clearing chemicals and is susceptible to the toxicity from these agents. Certain medicinal agents, when taken in overdoses and sometimes even when introduced within therapeutic ranges, may injure the organ. Chemicals that cause liver injury are called hepatotoxins (Friedman et al., 2003).

Hepatotoxicity is one of the main reasons behind withdrawal of a drug from the market. Fifty percent of all acute liver failures and 5% of all hospital admissions are associated with drug-induced hepatotoxicity (Dey et al., 2013).

A number of plants have been shown to possess hepatoprotective property by improving antioxidant status. Thus, the efficacy of the drug would be preventive and passive for defending against damages. Traditional medicines are effective in certain disorders and are based on experience in the use of plant products in amelioration of common diseases. Several Indian medicinal plants have in recent times been explored and the hepatoprotective effects of these plants have been established (Kumar et al., 2009; Abdel-Salam et al., 2014[1]).

Plant drugs are known to play a major role in the management of liver diseases. There are many plants and their extracts that have been shown to possess hepatoprotective activities (Girish and Pradhan, 2012).

Treatment options for common liver diseases are limited, and therapy with modern medicine may lack in efficacy. The effectiveness of treatments such as those using corticosteroids and interferons is inconsistent, carries the risk of adverse events, and is often too costly (Stickel and Schuppan, 2007). So, there is a need for effective therapeutic agents with a low incidence of side effects. The natural antioxidants, more recently, have attracted considerable attention of users and researchers largely on account of adverse toxicological reports on some synthetic antioxidants and growing awareness among consumers (Ramalakshmi et al., 2007). In fact, a single plant may have diversity of phytochemicals ranging from bitter compounds that stimulate digestive system, phenolic compounds for antioxidant and many other pharmacological properties, including antibacterial and antifungal, tannins that work as natural antibiotics, diuretic substances, and alkaloids. (Miguel, 2010).

2. The liver

The liver is the largest solid organ in the body, weighing about 1.5 kg in an adult. It lies in the right upper quadrant of the abdomen completely protected by the thoracic rib cage (Fig. 1). The liver is connected to two large blood vessels, one called hepatic artery and the other called the portal vein. The hepatic artery carries blood from the aorta whereas the portal vein carries blood containing digested food from the small intestine. The basic functional unit of the liver is the liver lobule; the human liver contains 50,000 to 100,000 individual lobules (Adi and Alturkmani, 2013).

2.1. *Hepatic physiology*

The liver is the center of metabolic homeostasis (Adi and Alturkmani, 2013; Sherwood, 1997). It serves as:

1. Vascular functions: for storage and filtration of blood.
   a. The liver can store 200–400 ml of blood in liver sinusoids (useful in hemorrhage).

[1] Underline (Abdel-Salam et al., 2014) means that this is Egyptian research.

b. Kupffer cells (highly phagocytic) can remove 90% of bacteria in the portal venous blood.
c. Hepatocytes synthesize plasma proteins.

2. Metabolic functions: the liver cells have a very high metabolic rate.
   a. Carbohydrate metabolism: the liver act as a glucostat under the effect of hormones (glycogenesis, glycogenolysis, gluconeogenesis).
      – Processing nutrients absorbed from digestive tract. The liver converts glucose into glycogen, its storage form. This glycogen can then be transformed back into glucose if the body needs energy (Guyton and Hall, 2006).
   b. Protein metabolism: deamination of amino acids, formation of urea, synthesis of 90% of plasma proteins and all non-essential amino acids.
      – Synthesizing plasma proteins, including those necessary for blood clotting. Most of the 12 clotting factors are plasma proteins produced by the liver. If the liver is damaged or diseased, it can take longer for the body to form clots. Other plasma proteins produced by the liver include albumin which binds many water-insoluble substances and contributes to osmotic pressure, fibrinogen which is the key to the clotting process, and certain globulins which transport substances such as cholesterol and iron.
      – Producing immune factors and removing bacteria, helping the body fight infection. The phagocytes in the liver produce acute-phase proteins in response to microbes. These proteins are associated with the inflammation process, tissue repair, and immune cell activities (Reichen, 1999).
   c. Fat metabolism: oxidation of fatty acids, formation of lipoproteins, cholesterol and phospholipids.
      – The fatty acids produced by the digestion of lipids are used to synthesize cholesterol and other substances. The liver also has the ability to convert certain amino acids into others (Guyton and Hall, 2006).
   d. Storage of vitamins: (A, K, B12, and D) and iron
      – Storing certain vitamins, minerals, and sugars. The liver stores enough glucose in the form of glycogen to provide about a day's worth of energy. The liver also stores fats, iron, copper, and many vitamins including vitamins A, D, K, and B_{12} (Adi and Alturkmani, 2013; Sherwood, 1997).
   e. Detoxification: or excretion of drugs, hormones and other substances.
      – Removing and excreting body wastes and hormones as well as drugs and other foreign substances. These substances have entered the blood supply either through production by metabolism within the body or from the outside in the form of drugs or other foreign compounds. Enzymes in the liver alter some toxins so they can be more easily excreted in urine.
      – Excretion of bilirubin. Bilirubin is one of the few waste products excreted in the bile. Macrophages in the liver remove worn out red blood cells from the blood. Bilirubin then results from the breakdown of the hemoglobin in the red blood cells and is excreted into the bile by hepatocytes. Jaundice results when bilirubin cannot be removed from the blood quickly enough due to gallstones, liver disease, or the excessive breakdown of red blood cells (Adi and Alturkmani, 2013; Sherwood, 1997).

3. Secretory and excretory function: formation of bile.
   – Producing bile to aid in digestion. Bile salts aid in fat digestion and absorption. Bile is continuously secreted by the liver and stored in the gallbladder until a meal, when bile enters the beginning of the small intestine. Bile production ranges from 250 ml to 1 l per day depending on the amount of food eaten (Guyton and Hall, 2006).

2.2. *Physiological changes due to hepatotoxicity*

The liver plays a central role in transforming and clearing chemicals and is susceptible to the toxicity from these agents. More than 900 drugs have been implicated in causing liver injury (Friedman et al., 2003) and it is the most common reason for a drug to be withdrawn from the market. Hepatotoxicity and drug-induced liver injury also account for a substantial number of compound failures, highlighting the need for drug screening assays, such as stem cell-derived hepatocyte-like cells, that are capable of detecting toxicity early in the drug development process (Greenhough et al., 2010). Chemicals often cause subclinical injury to the liver, which manifests only as abnormal liver enzyme tests. Drug-induced liver injury is responsible for 5% of all hospital admissions and 50% of all acute liver failures (Nally and Peter, 2006; Ostapowicz et al., 2002).

2.3. *Drug metabolism in the liver*

The human body identifies almost all drugs as foreign substances (i.e. xenobiotics) and subjects them to various chemical processes (i.e. metabolism) to make them suitable for elimination. This involves chemical transformations to reduce fat solubility and to change biological activity. Although almost all tissues in the body have some ability to metabolize chemicals, smooth endoplasmic reticulum in the liver is the principal "metabolic clearing house" for both endogenous chemicals (e.g., cholesterol, steroid hormones, fatty acids, proteins) and exogenous substances (e.g., drugs, alcohol) (Blumenthal et al., 2006).

Drug metabolism is usually divided into two phases: phase 1 and phase 2. Phase 1 reaction is thought to prepare a drug for phase 2. However, many compounds can be metabolized by phase 2 directly. Phase 1 reaction involves oxidation, reduction, hydrolysis, hydration and many other rare chemical reactions. These processes tend to increase water solubility of the drug and can generate metabolites that are more chemically active and potentially toxic. Most of phase 2 reactions take place in cytosol and involve conjugation with endogenous compounds via transferase enzymes. A group of enzymes located in the endoplasmic reticulum, known as cytochrome P-450, is the most important family of metabolizing enzymes in the liver. Cytochrome P-450 is the terminal oxidase component of an electron transport chain. It is not a single enzyme, but rather consists of a closely related family of 50 isoforms; six of them metabolize 90% of drugs. There is a tremendous diversity of individual P-450 gene products, and this heterogeneity allows

the liver to perform oxidation on a vast array of chemicals (including almost all drugs) in phase 1 (Lynch and Price, 2007).

2.4. *Mechanism of liver damage*

Drugs continue to be taken off the market due to late discovery of hepatotoxicity. Due to its unique metabolism and close relationship with the gastrointestinal tract, the liver is susceptible to injury from drugs and other substances. Seventy five percent of blood coming to the liver arrives directly from gastrointestinal organs and then spleen *via* portal veins that bring drugs and xenobiotics in near-undiluted form. Several mechanisms are responsible for either inducing hepatic injury or worsening the damage process. Many chemicals damage mitochondria, an intracellular organelle that produces energy. Its dysfunction releases excessive amount of oxidants that, in turn, injure hepatic cells (Fig. 2). Activation of some enzymes in the cytochrome P-450 system such as CYP2E1 also leads to oxidative stress, which in turn has a crucial role in liver damage. Injury to hepatocyte and bile duct cells leads to accumulation of bile acid inside the liver. This promotes further live damage (Jaeschke et al., 2002).

2.5. *Patterns of injury*

Chemicals produce a wide variety of clinical and pathological hepatic injury. Biochemical markers (e.g. alanine transferase, alkaline phosphatase and bilirubin) are often used to indicate liver damage. Liver injury is defined as a rise in either (a) ALT level more than three times of the upper limit of normal (ULN), (b) alkaline phospatase (ALP) level more than twice ULN, or (c) total bilirubin level more than twice ULN when associated with increased ALT or ALP (Mumoli et al., 2006). Liver damage is further characterized into hepatocellular (predominantly initial alanine transferase elevation) and cholestatic (initial alkaline phosphatase rise) types. However, they are not mutually exclusive and mixed types of injuries are often encountered. The liver synthesizes, concentrates, and secretes bile acids and excretes other toxicants, such as bilirubin. Drug-induced injury to hepatocytes and bile duct cells can lead to cholestasis. Cholestasis, in turn, causes intrahepatic accumulation of toxic bile acids and excretion products, which promotes further hepatic injury (Jaeschke et al., 2002).

Marker enzymes are widely used to assess liver damage; hence, we determined the levels of enzymes such as ALP, AST and ALT. Necrosis or membrane damage releases the enzyme into circulation; hence, it can be measured in the serum. High levels of AST indicate liver damage, such as that caused by viral hepatitis as well as cardiac infarction and muscle injury; AST catalyzes the conversion of alanine to pyruvate and glutamate and is released in a similar manner. Therefore, ALT is more specific to the liver, and is thus a better parameter for detecting liver injury. Elevated levels of serum enzymes are indicative of cellular leakage and loss of functional integrity of cell membrane in liver (Choudhary and Devi, 2014). Serum ALP, bilirubin and total protein levels, on the other hand, are related to the

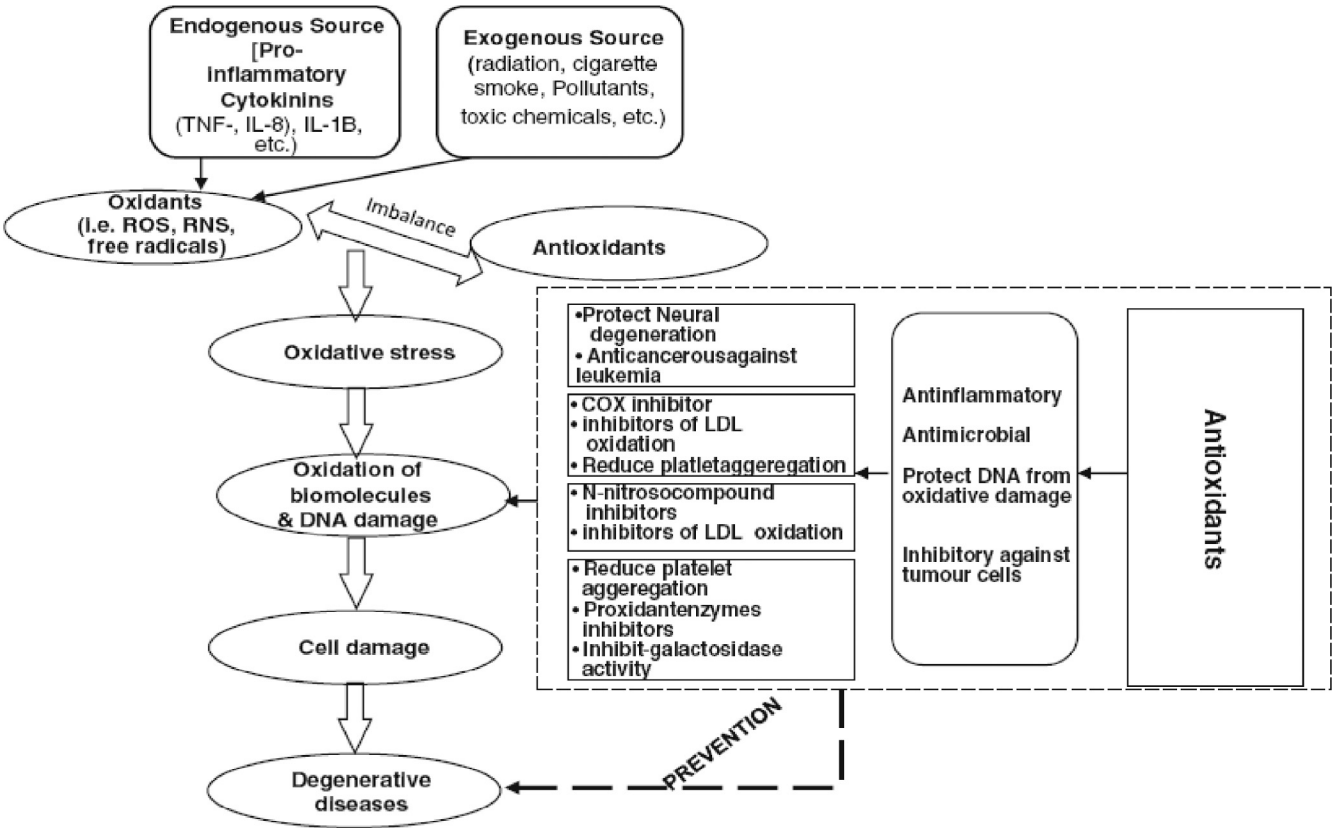


Fig. 2 – Potential of antioxidants in preventing oxidation of biomolecules, DNA damage and degenerative diseases (openi.nlm.nih.gov).

function of hepatic cell. Increase in serum level of ALP is due to increased synthesis, in the presence of increasing biliary pressure (Muriel and Garcipiana, 1992). Increase in the level of these serum enzymes ALP, acid phosphatase (ACP), AST, ALT, lactate dehydrogenase (LDH) and gamma-glutamyl transferase (γ-GT) was also observed. Lipid peroxidation leads to generation of free radicals (such as peroxyl, alkoxyl and aldehyde) that cause cell damage and leads to the release of marker enzymes. Disruption of the ordered lipid-bilayer of the membrane structure is probably due to the presence of reactive oxygen species produced due to oxidative stress, leading to the escape of detectable quantity of these enzymes out of the cell into the extracellular fluid. The reactive oxygen species might have oxidized the polyunsaturated fatty acids which make up the lipid bilayer resulting in its disruption. The elevated levels of serum globulin also suggest adverse effect of reactive oxygen species on the secretory ability of the liver, and hence it also affects the normal functioning of the organ. Increased bilirubin production, enhanced hepatic conjugation and biliary excretion of the pigment present in aspartame-treated animals may be a result of decreased uptake, conjugation or increased bilirubin production (Choudhary and Devi, 2014).

2.6. *Chemicals and drug-induced hepatotoxicity*

Aspartame is widely consumed by humans who are diabetic and who are under weight loss regime. Aspartame (L-aspartyl-L-phenylalanine methyl ester) also known as NutraSweet, after oral administration to humans and experimental animals, is rapidly and completely metabolized to 50% phenylalanine, 40% aspartic acid and 10% methanol. Methanol is being increasingly recognized as a substance that damages the liver cells, where it is oxidized to formaldehyde and later to format (Oppermann, 1984). These processes are accompanied by elevation of NADH level and the formation of superoxide anion, which may be involved in lipid peroxidation (Parthasarathy et al., 2006). Also, methanol intoxication is associated with mitochondrial damage and increased microsomal proliferation, resulting in increased production of oxygen radicals (Castro et al., 2002). These factors together with the excess of formaldehyde formed during acute methanol intoxication cause significant increase in lipid peroxidation (Parthasarathy et al., 2006). Aspartame consumption in a long-term basis may affect the brain and liver, and it may be due to its metabolite methanol or aspartame may act as a chemical stressor to alter the antioxidant status and histological pattern. Also, aspartame can induce oxidative stress and hepatorenal toxicity. Long-term treatment with aspartame may be responsible for oxidative stress and hepatorenal toxicity (Ashok et al., 2013).

Carbon tetrachloride (CCl_4) has been widely used for experimental induction of liver damage (Parola et al., 1992). The principal causes of carbon tetrachloride (CCl_4)-induced hepatic damage are lipid peroxidation and decreased activities of antioxidant enzymes and generation of free radicals (Poli, 1993).

Doxorubicin (DXR, trade name Adriamycin; also known as hydroxy daunorubicin) has been widely used over the past several decades to treat patients with various cancers, including hepatocellular carcinoma, based on its ability to kill transformed liver cells (Singal and Iliskovic, 2008). Liver damage is a relatively common adverse effect in patients with other cancers who are treated with DXR (Cainelli and Vallone, 2009). DXR hepatotoxicity has been also reported in a number of studies on animals (Ray et al., 2000; Ray and Mehendale, 2000; Kalender et al., 2001; Ray, 2003). The mechanisms of DXR cytotoxicity in cancer cells include (i) intercalation into DNA with inhibition of DNA replication and RNA transcription; (ii) generation of free radicals with DNA damage and lipid peroxidation; (iii) DNA binding and arylation; (iv) DNA crosslinking; (v) interference with DNA unwinding, DNA strand separation, and helicase activity; (vi) direct membrane damage due to oxidation of lipids; and (vii) inhibition of topoisomerase II (Zhang et al., 2009).

Acetaminophen (APAP) is the most common non-steroidal analgesic and antipyretic drug used worldwide. APAP exerts few side effects in therapeutic doses, but hepatotoxicity is the frequent consequence of APAP overdose (Olaleye et al., 2010). APAP overdose is responsible for generation of a highly reactive metabolite N-acetyl-p-benzoquinoneimine (NAPQI). NAPQI causes covalent modification, reduction of enzyme activity, inhibition of protein oxidation, free radical generation, lipid peroxidation, DNA fragmentation, mitochondrial dysfunction, alteration of innate immunity, deregulation of Ca2+ homeostasis, and depletion of glutathione, thus leading to hepatotoxicity (James et al., 2003).

2.7. *Oxidative stress effect on hepatotoxicity*

Oxidative stress reflects an imbalance between the systemic manifestation of reactive oxygen species and a biological system's ability to readily detoxify the reactive intermediates or to repair the resulting damage. Disturbances in the normal redox state of cells can cause toxic effects through the production of peroxides and free radicals that damage all components of the cell, including proteins, lipids, and DNA. Oxidative stress from oxidative metabolism causes base damage, as well as strand breaks in DNA. Base damage is mostly indirect and caused by the reactive oxygen species (ROS) generated, e.g. O_2– (superoxide radical), OH (hydroxyl radical) and H_2O_2 "hydrogen peroxide" (Chandra et al., 2015).

Oxidative stress affects the major cellular components: proteins, lipids and DNA. The importance of oxidative stress is commonly emphasized in the pathogenesis of various degenerative diseases, such as diabetes, cancer, cardiovascular disorders or neurodegenerative diseases (Apostolova et al., 2011). Hence, the role of oxidant agents in cells is complex and depends on the balance between oxidant and antioxidant particles. Protective actions against ROS are performed by several enzymes [(e.g., superoxide dismutase (SOD), catalase and glutathione peroxidase)] as well as nonenzymatic compounds (e.g., tocopherol, vitamin E, beta-carotene, ascorbate and glutathione (GSH)) (Mao et al., 2011). When the capacity of this antioxidant system decreases, the level of inactivated ROS rises. ROS generation also leads to altered mitochondrial permeability and transition potential. These changes induce the release of pro-apoptotic factors (e.g., cytochrome C) (Wang et al., 2013). Additionally, oxidative stress may induce reversible and irreversible changes in sensitive proteins. This process is often associated with neurodegenerative disorders (Lu, 2008).

2.8. *The role of lipid peroxidation in hepatotoxicity*

Lipid hydroperoxides are non-radical intermediates derived from unsaturated fatty acids, phospholipids, glycolipids, cholesterol esters and cholesterol itself. Their formation occurs in enzymatic or non-enzymatic reactions involving activated chemical species known as "reactive oxygen species" (ROS), which are responsible for toxic effects in the body *via* various tissue damages. These ROS include among others hydroxyl radicals, lipid oxyl or peroxyl radicals, singlet oxygen, and peroxynitrite formed from nitrogen oxide (NO), all these groups of atoms behave as a unit and are now named "free radical." Lipid peroxidation leads to generation of free radicals (such as peroxyl, alkoxyl and aldehyde), which cause cell damage and lead to the release of marker enzymes. When liver and kidney cell plasma membrane is damaged, a variety of enzymes normally located in the cytosol are released in to the blood stream. Their estimation in the serum is a useful quantitative marker of the extent and type of hepatic and renal cellular damage (Pagana and Pagana, 2002).

3. Protective role of herbal plants

3.1. *History*

The study of herbs dates back over 5000 years to the Sumerians, who described well-established medicinal uses for such plants as laurel, caraway and thyme. Ancient Egyptian of 1000 B.C. are known to have used garlic, opium, castor oil, coriander, mint, indigo, and other herbs for medicine, and the old testament also mentioned herb use and cultivation, including mandrake, vetch, caraway, wheat, barley and rye. Also, the use of herbal drugs can be traced back to 2100 B.C. in ancient China at the time of Xia dynasty and in India during Vedic period. The first written report dates back to 600 B.C. with the Charaka Samhita of India and the early notes of Eastern Zhou dynasty of China, which became systematized around 400 B.C. The age-old system of herbal medicine is being revived by day-to-day practice for its long-lasting curative effect, easy availability, natural way of healing, and less side effects (Schuppan et al., 1999).

3.2. *Plants used as a medicine*

Herbal medicines are gaining importance not only in India, but all over the world. Alternate system of medicine in India uses about 1200 plants for different ailments (Rai, 1994). Treatment options for common liver diseases are limited and therapy with modern medicine may lack in efficacy. The effectiveness of treatments such as those using corticosteroids and interferons is inconsistent, carries the risk of adverse events and is often too costly (Stickel and Schuppan, 2007). So, there is a need for effective therapeutic agents with a low incidence of side effects. Plants potentially constitute such a group, which is traditionally used as hepatoprotective agents (Luper, 1998). The easy accessibility without the need of laborious pharmaceutical synthesis increased the attention toward the herbal medicines (Stickel and Schuppan, 2007). Plant drugs are also known to play a major role in the management of liver diseases all over the world. There are many plants and herbal extracts that have been shown to possess hepatoprotective activities in Indian, Chinese, and Korean system of medicine (Thyagarajan et al., 2002).

3.3. *Plants biosynthesize a variety of phytochemicals*

Phytochemicals are a large group of plant-derived compounds hypothesized to be responsible for much of the disease protection conferred from diets high in fruits, vegetables, beans, cereals, and plant-based beverages such as tea and wine (Arts and Hollman, 2005).

Phytochemicals include the following:

Alkaloids contain a ring with nitrogen. Many alkaloids have dramatic effects on the central nervous system. Caffeine is an alkaloid that provides a mild lift, but the alkaloids in Datura cause severe intoxication.

Phenolic contains phenol ring. The anthocyanins that give grapes their purple color, the isoflavones, the phytoestrogens from soy, and the tannins that give tea its astringency are phenolics (Table 1).

Terpenoids are built up from terpene building blocks. Each terpene consists of two paired isoprenes. The names monoterpenes, sesquiterpenes, diterpenes and triterpenes are based on the number of isoprene units. The fragrance of rose and lavender is due to monoterpenes. The carotenoids produce the reds, yellows and oranges of pumpkin, corn and tomatoes.

Glycosides consist of glucose moiety attached to an aglycone. The aglycone is a molecule that is bioactive in its free form but inert until the glycoside bond is broken by water or enzymes.

The mechanism of hepatoprotection by these compounds generally exerts multiple effects. Although they show hepatoprotection due to antioxidant effect, there are other effects like immunomodulatory, antiviral (Wiart et al., 2005) and anti-inflammatory (Sandur et al., 2007).

The use of herbal resources for the treatment of liver diseases is quite an old approach of various traditional systems of medicine. These medicine systems conceptualize a general imbalance of the dichotomous energies that leads to the disease and they focus on medicine that balances these energies and maintains good health. Mainly, herbs have been used for chronic hepatitis C and alcohol-induced liver diseases. Phytomedicines are traditionally used in the treatment of liver disorders and are now included as complementary and alternative medicine for liver patients. Several hundred plants have been examined for use in a wide variety of liver disorders (Negi et al., 2008).

3.3.1. Zingiber officinale *(ginger)*

Ginger (*Zingiber officinale* Roscoe, Zingiberaceae) (Fig. 3) is widely used around the world in foods as a spice. For centuries, it has been an important ingredient in Chinese, Ayurvedic and Tibb-Unani herbal medicines for the treatment of catarrh, rheumatism, nervous diseases, gingivitis, toothache, asthma, stroke, constipation and diabetes (Awang, 1992; Tapsell et al.,

Table 1 – Examples of some flavonoids and their food sources (Lakhanpal and Rai, 2007).

Groups	Compounds	Food sources
Flavonols	Quercetin Kaempferol Isorhamnetin Quercetagetin	Yellow onion, curly kale, leek, cherry tomato, broccoli, apple, green and black grapes, blueberry
Flavones	Tangeretin Heptamethoxyflavone Nobiletin Sinensetin Quercetogetin Chrysin Luteolin Disometin Tricetin	Parsley, celery, Capsicum pepper
Flavanones	Naringenin Eriodictyol Hesperetin Dihydroquercetin Dihydrofisetin Dihydrobinetin	Orange juice, grapefruit juice, lemon juice
Flavanols	Silibinin Silymarin Taxifolin Pinobanksin	Coca, cocoa beverages, chocolates
Catechins (proanthocyanidins)	(+) Catechin Gallocatechin (–) Epicatechin Epigallocatechin Epicatechin 3-gallate Epigallocatechin 3-gallate	Chocolate, beans, apricot, cherry, grapes, peach, red wine, cider, green tea, black tea, blackberry
Isoflavones	Daidzein Genistein Glycitein	Soy, cheese, soy flour, soy bean, tofu
Anthocyanins	Cyanidin Delphinidin Malvidin Pelargonidin Peonidin Petunidin	Blueberry, blackcurrant, black grapes, cherry, rhubarb, plum, strawberry, red wine, red cabbage

2006; Wang and Wang, 2005). Among the pharmacological effects demonstrated are anti-platelets, antioxidant, anti-tumor, anti-rhino viral, anti-hepatotoxicity, anti-arthritic and anti-inflammatory (Lantz et al., 2007). The antioxidant activity of gingerol and other constituents of ginger has been confirmed (Aeschbach et al., 1994). Different doses of ginger extract cause alterations in biochemical parameters, free radicals, antioxidant enzymes and drug metabolizing enzymes induced by bromobenzene in the liver of male rats and alleviating the toxicity of bromobenzene in the liver (El-Sharaky et al., 2009). Curcumin, another active component present in ginger, was found to be an antioxidant and anti-inflammatory agent and induced haem oxygenase-1 and protected endothelial cells against oxidative stress (Motterlini et al., 2000). The antioxidants inhibit the reactive oxygen species (ROS), which are capable of causing damage to DNA, associated with carcinogenesis, coronary heart disease, and many other health problems related to advancing age (Patel et al., 2000). The aqueous extract of ginger root may cause hepatoprotective effect against aspartame, which may cause hepatotoxicity and oxidative stress. Ginger root extract has hepatoprotective effect against aspartame-induced hepatotoxicity and decreased liver function markers (ALT, AST, ALP, γ-GT), serum total protein, albumin and total bilirubin levels, serum LDH activity, α-fetoprotein, and tumor necrosis factor (TNF), increased levels of antioxidant enzymes, and reduced levels of malondialdehyde (Hozayen and Abou Seif, 2014).

Fig. 3 – *Zingiber officinale* (http://www.alvita.com/herbal-teas.html).

Fig. 4 – *Cucurbita pepo L* (Various, 2013).

3.3.2. Cucurbita pepo L. *(pumpkin)*

Pumpkin seeds (*Cucurbita pepo L.*) (Fig. 4) are a rich source of unsaturated fatty acids, antioxidants and fibers, known to have anti-atherogenic and hepatoprotective activities (Makni et al., 2008). Pumpkin is one such plant that has been frequently used as functional food or medicine (Caili et al., 2006). Some of its common uses in most countries are for diabetes where it is used internally as well as externally for management of worms and parasites. Treatment of spontaneously hypertensive rats with felodipine or captopril monotherapy or combined with pumpkin seed oil produced improvement in the measured free radical scavengers in the heart and kidney (Al-Zuhair et al., 2000). Pumpkin is also rich in unsaturated fatty acids especially linoleic and oleic acid and tocopherols and with very high oxidative stability (Stevenson et al., 2007). In addition to the carotenoids and gamma aminobutyric acids (GABA) found in the fruits (Liu et al., 2001), there are other biologically active ingredients, which are found in pumpkins (Gossell-Williams et al., 2008), such as sterols, proteins, peptides, polysaccharides, para-aminobenzoic acid and fixed oils. Pumpkin seed oil's main nutrients are: essential fatty acid-omega 6, omega 9, phytosterols, and antioxidants such as carotenoids, vitamin A and vitamin E (Murkovic et al., 1996). Linoleic acid, a polyunsaturated fatty acid present in pumpkin seed oil, is known to increase membrane fluidity and allows for osmosis, intracellular and extracellular gaseous exchange (Lovejoy, 2002). Pumpkin seed oil includes fatty acids: palmitic (C 16:0), stearic (C 18:0), oleic (C 18:1) and linoleic (C 18:2) (Kulaitiene et al., 2007). Antioxidants are the substances that when present in low concentration significantly delay or reduce the oxidation of the substrate (Halliwell, 2000). Pumpkin oil may play an important role in the protection against alcohol-induced hepatotoxicity and oxidative stress. Pretreatment with pumpkin oil may have hepatoprotective effects, which are varied and include oxidation, anti-lipid peroxidation enhanced detoxification and protection against glutathione depletion (Abou Seif, 2014a).

3.3.3. Citrus reticulata *(mandarin)*

The mandarin (*Citrus reticulata*), also known as the mandarine (Fig. 5), is a small citrus tree with fruit resembling other oranges. They are rich in vitamin C, flavonoids, acids and volatile oils. Mandarin, as other citrus fruits, has nutritional importance due to its particular composition. Flavonoids, especially polymethoxy flavones and flavanones (hesperidin, rutin and naringin), are identified in citrus pulp as well as in peel (Wang et al., 2008).

Rutin (quercetin rutinoside) is a glycoside of the flavonoid quercetin. Rutin is used as a medication for blood vessel protection and is an ingredient of numerous multivitamin preparations and herbal remedies. It can combine with cations, supplying nutrients from the soil to the cells in plants (Luo et al., 2008). In humans it is a potent antioxidant where its actions include attaching to the iron ion (Fe^{2}+), and preventing it from binding to hydrogen peroxide, which would otherwise create a highly reactive free-radical that may damage cells (Patel et al.,

Fig. 5 – *Citrus reticulate* (Brown, 2002).

Fig. 6 – *Petroselinum crispum* (www.edenbrothers.com).

Fig. 7 – *Andrographis paniculata* (Zell, 2009).

2010). Rutin is the strongest antioxidant when compared with quercetin, acacetin, morin, hispidulin, hesperidin, and naringin (Chow et al., 2005), which are mostly isolated from citrus peel.

Hesperidin is a flavanone glycoside (flavonoid $C_{28}H_{34}O_{15}$) found abundantly in citrus fruits. Its aglycone form is called hesperetin. Hesperidin is believed to play a role in plant defense. It acts as an antioxidant according to in vitro studies. In human nutrition, it contributes to the integrity of blood vessels (Farombi et al., 2008). Hesperidin also has anti-inflammatory effects (Emim et al., 1994). Dietary hesperidin exerts anti-carcinogenic actions in the tongue, colon, esophagus, and urinary bladder in rat carcinogenesis models (Tanaka et al., 2000). Hesperidin has been reported to have many biological effects, including anti-inflammatory, antimicrobial, anticarcinogenic, antioxidant effects and decreasing capillary fragility (Garg et al., 2001). Hesperidin, a flavonoid, has powerful protection effects on DNA damage, reducing the frequency of micronuclei induced by c-irradiation in mice (Hosseinimehr and Nemati, 2006). Hesperidin plays an important role in the protection against doxorubicin-induced hepatotoxicity, by improving the activities of liver enzymes (ALT, AST, and ALP and GGT) in addition to the amelioration in the levels of total bilirubin, albumin and sialic acid. Rutin and hesperidin recorded a significant increase in the liver glutathione level, glutathione peroxidase, glutathione-S-transferase and peroxidase activities, and reduced lipid peroxidation level. Pretreatment with rutin and hesperidin may protect the liver from the hepatotoxic effect caused by doxorubicin (Hozayen et al., 2014).

3.3.4. Petroselinum crispum *oil (parsley oil)*

Parsley, *Petroselinum crispum* (Fig. 6), is a native herb of the central Mediterranean region (southern Italy, Algeria, and Tunisia). It is a part of the Apiaceae family and is a species of *Petroselinum* (Lopez et al., 1999). The phenolic compounds of parsley were responsible for its antibacterial and antioxidant activity (Wong and Kitts, 2006). The antioxidant activity of this herb in terms of B-carotene possesses bleaching capacity and free radical scavenging activity (Zhang et al., 2006). This concept was then confirmed by further studies (Kolarovic et al., 2010). Parsley possesses several flavonoids such as apiin and luteolin, and its essential oil contains apiol and myristicin. These components are believed to be responsible for the therapeutic effects of parsley (Mimica-Dukic and Popovic, 2007). Parsley oil plays an important role by exerting an ameliorating effect on liver function enzymes, antioxidation, anti-lipid peroxidation that enhanced detoxification, and protection against glutathione depletion against alcohol-induced hepatotoxicity and oxidative stress (Abou Seif, 2014b).

3.3.5. Andrographis paniculata *(king of bitters)*

Andrographis paniculata (*A. paniculata*), known among the Indians, is one of the most commonly used plants in the traditional systems of Unani and Ayurvedic medicines. It is called Creat in English and is known as the king of bitters (Jarukamjorn and Nemoto, 2008). The aerial parts (Fig. 7) are most commonly used; however, the whole plant or the roots are also used for certain purposes in some manuscripts. *A. paniculata* has been reported as having antibacterial (Leelarasamee et al., 1990), anti malarial (Dua et al., 2004), antiviral (Wiart et al., 2005), cardioprotective, antioxidant, anti-inflammatory (Sheeja et al., 2006), antidiabetic effects and also antitumor activities (Zhao et al., 2008). Treatment with whole plant extract of *A. paniculata* effectively reduced the level of lipid peroxidation and increased the status of antioxidant enzymes. This may be due to the presence of various flavonoids, phenols and glycosides in the drug (Subramaniam et al., 2015).

3.3.6. Silybum marianum *(milk thistle)*

Silymarin, derived from the seeds of *Silybum marianum L.* (Family: Asteraceae or Compositae), is a member of sunflower family and commonly called milk thistle. The plant has been used for centuries as a natural remedy for liver and biliary tract diseases. The leaves are characterized by distinct white "milky" veins that give the plant its common name. The active constituents of thistle are flavonolignans, including silybin, silydianin and silychristin, collectively known as silymarins (Shaarawy et al., 2009). Silybin is the component with the greatest degree of biologically active and milk thistle extracts are usually standardized to contain 70–80 percent silybin. Silymarin is found in the entire plant but is concentrated in the fruit (Fig. 8) and seeds. Silybum seeds also contain betaine (a proven hepatoprotector) and essential fatty acids, which may contribute to silymarins' anti-inflammatory effect (Saller et al., 2001).

Silymarin has been reported to protect liver cells from a wide variety of toxins, including acetaminophen, ethanol, CCl_4 and D-galactosamine (Rasool et al., 2014). The mechanisms that provide silymarins their hepatoprotective effects are many and varied, and include anti-oxidation, anti-lipid peroxidation enhanced detoxification and protection against glutathione depletion (Pradhan and Girish, 2006).

Fig. 8 – *Silybum marianum* (www.amazon.com).

3.3.7. Camellia sinensis *(green tea)*

Green tea leaves (Fig. 9) produce organic compounds that may be involved in the defense of plants against invading pathogens, and these metabolites are known as polyphenols (Friedman, 2007), which include catechin, epicatechin, epigallocatechin, tannins and caffeine (Wang and Goodman, 1999). Green tea displays antioxidants and free radical scavenger properties (Crespy and Williamson, 2004). The green tea extract and its main catechin polyphenols have medicinal value for the prevention of and therapeutics in several diseases (Ostrowska and Skrzydlewska, 2006). The green tea exerts improvement in liver function by preventing the production of reactive oxygen species (ROS) and enhancing the antioxidant defense system capacity. Thus green tea extract has protective effects against ethanol toxicity (Lodhi et al., 2014).

3.4. *Drugs derived from herbal plants*

Many drugs are derived from plants. Some examples of drugs that consist of plant ingredients are shown in Table 2 and include the following:

Fig. 9 – *Camellia sinensis* (www.camelliasrus.com.au).

a. **Lovenox**: The active principles: curcumin 2%, silymarin 80% and dandelion extract (*Taraxacum officinale*). It improves liver functions.
b. **Hepanox capsules**: It contains silymarin and vitamins A, C and E. It is hepatoprotective and antioxidant.
c. **Legalon**: The main active principle is silymarin, which improves liver function and protects against liver damage.
d. **Lipocholine tablets: lipotropic agent**: The active constituents are extract of artichoke (*Cynara scolymus*) and a group of vitamin B.

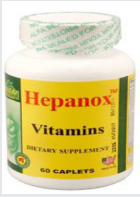
4. Summary and conclusion

The liver is the largest, important organ and the site for essential biochemical reactions in the human body. It has the function to detoxify toxic substances and synthesize useful biomolecules. Therefore, damage to the liver leads to grave consequences. This damage resulted from chronic alcoholic abuse, viral hepatitis or inherited metabolic disease. Liver damage is associated with cellular necrosis, fibrosis, and increase in tissue lipid peroxidation and depletion in tissue glutathione level. Most of the hepatotoxic chemicals damage liver cells mainly by inducing lipid peroxidation and other oxidative damages.

Natural antioxidants are found in many compounds classified as secondary plant metabolites, e.g. polyphenols (phenolic acids and flavonoids) and terpenoids (carotenoids), and the consumption of foods that contain these compounds in large quantities seems to play an important role in prophylaxis against many diseases.

Herbal medicines derived from plant extracts are being increasingly utilized to treat a wide variety of clinical disease. More attention has been paid to the protective effects of natural antioxidants against drug-induced toxicities especially whenever free radical generation is involved. Popularity of herbal remedies is increasing and at least one quarter of patients with liver disease use botanicals. The World Health Organization (WHO) estimates that 80 percent of the population of some Asian and African countries presently use herbal medicine for some aspects of primary health care. Some medicinal herbs have proven hepatoprotective potential. *Silybum marianum* (milk thistle) has been used to treat liver diseases since the 16th century. Its major constituents are the flavonoids silibinin, silydianin, silychristin, and isosilibinin, of which silibinin is the biologically most active compound and used for standardization of pharmaceutical products.

Table 2 – Drugs derived from herbal plants.

Lovenox	Hepanox capsules	Legalon	Lipocholine tablets
			
www.suggestkeyword.com	www.thevits.com	goldpharma.com	www.solgaronline.co.uk

4.1. *Egyptian experience*

Medicinal plants have been used as a source of remedies since ancient times in Egypt. Egyptian experience was previously mentioned inside the literature (underlined authors), such as Al-Zuhair et al. (2000), Abdel-Salam et al. (2014), Abou Seif (2014a,b), Hozayen and Abou Seif (2014, 2015), Hozayen et al. (2014), and Shaarawy et al. (2009).

5. Recommendation

Popularity of herbal remedies is increasing and at least one quarter of patients with liver disease use botanicals.

- Further collaboration between researchers in the fields of biology, pharmacognosy and medicinal sciences is required to elucidate the biological activity of the medicinal herbs' constituents and to assess their efficacy and safety in human beings and approval by the Food and Drug Administration (FDA).
- Egypt has a great role to play, as supplier of herbal products, not only to meet the domestic needs, but also to take advantage of the tremendous export potential.
- In Egypt, all herbal ingredients must be classified according to their botanical names beside their popular/common (commercial) names.
- Focusing on such phytochemicals and its cytotoxic mechanism might be valuable in determining the role of these chemicals in normal and altered body physiology.
- Daily foods must contain vegetables and fruits, which have polyphenolic compounds and vitamins that help to restore the balance between the antioxidants and free radicals and enhance body defense against diseases.

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